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Personalized medicine in metastatic non-small-cell lung cancer: promising targets and current clinical trials

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ABSTRACT

Non-small-cell lung cancer (NSCLC) remains the leading cause of cancer-related death globally, with most patients presenting with non-curable disease. Platinum-based doublet chemotherapy has been the cornerstone of treatment for patients with advanced-stage disease and has resulted in a modest increase in overall survival (on the order of an incremental 2 months increased survival per decade) and quality of life. Improved knowledge of the molecular signalling pathways found in NSCLC has led to the development of biomarkers with associated targeted therapeutics, thus changing the treatment paradigm for many NSCLC patients. In this review, we present a summary of many of the currently investigated NSCLC targets, discuss their current clinical trial status, and provide commentary as to the likelihood of their success making a positive impact for NSCLC patients.

KEY WORDS

Lung cancer, clinical trials, novel targets, novel therapeutics

1. INTRODUCTION

The global burden of non-small-cell lung carcinoma (NSCLC) and the very modest improvement in survival for advanced-stage patients since the early 1990s argues strongly for a paradigm shift in treatment strategies [1,2]. With the advent of improved molecular methodologies and, now, next-generation sequencing, it is predicted that during the next 5 years, most patients with NSCLC will be found to have either activating mutations, translocations creating fusion proteins, or gene amplifications that will allow for therapy with targeted agents or, at the very least, for the generation of treatment algorithms based on tumour re-classification prognostication [3].

There is little doubt that targeted therapies in NSCLC have increased survival in select patient groups. The small-molecule epidermal growth factor receptor (EGFR) tyrosine kinase inhibitors (TKIs) erlotinib and gefitinib now have defined roles in patient treatment.

Two pivotal phase III studies (IPASS and INTEREST) highlighted the role of gefitinib. In chemotherapy-naïve patients, IPASS compared gefitinib with carboplatin–paclitaxel. In the *EGFR*-unselected population, the study showed no benefit of gefitinib in overall survival, time to progression, or overall response rate. However, in patients with *EGFR* mutated tumours, progression-free survival (PFS) was significantly longer [hazard ratio (HR): 0.48; 95% confidence interval (CI): 0.36 to 0.64; $p < 0.001$] [4]. In the phase III INTEREST trial, single-agent gefitinib was noninferior to docetaxel in second- and third-line treatment. No difference in benefit was seen in patients with *EGFR* gene amplification; however, a suggestion of benefit in terms of overall response rate and PFS was observed in an unplanned analysis of patients with *EGFR* mutation [5].

The first phase III study directly comparing erlotinib with standard chemotherapy in the first-line advanced setting in Chinese patients with an activating *EGFR* mutation was the OPTIMAL trial. That trial showed a PFS of 13.1 months with erlotinib compared with 4.6 months with gemcitabine–carboplatin chemotherapy (HR: 0.16; 95% CI: 0.1 to 0.26; $p < 0.001$) [6]. A second trial called EURTAC, the first to involve a Western European population, randomized patients to a platinum-based doublet chemotherapy regimen (docetaxel–gemcitabine) or to erlotinib in patients with an *EGFR* activating mutation. Patients treated with erlotinib experienced a PFS advantage (9.7 months vs. 5.2 months; HR: 0.37; 95% CI: 0.25 to 0.54) [7]. Additionally, the phase III SATURN trial examined erlotinib as maintenance therapy after platinum-based chemotherapy. That trial met the primary endpoint of significantly longer PFS in patients treated with erlotinib (12.3 weeks) than in patients receiving placebo (11.1 weeks; HR: 0.69; 95%CI: 0.58 to 0.82; $p < 0.0001$) [8].

Similarly, the roles of *EML4–ALK* gene rearrangements and of targeted Alk tyrosine kinase inhibitors as active agents in NSCLC patients have been established. Rearrangements of the *ALK* gene are felt to be mutually exclusive of *EGFR* and *KRAS* mutations and occur in approximately 4% of lung cancers. The *ALK* mutations are more common in adenocarcinomas and in light smokers or nonsmokers [9]. Crizotinib, an oral ATP-selective inhibitor of Alk tyrosine kinase, received approval from the U.S. Food and Drug Administration for that setting in 2011. The phase I trial of this agent in advanced *ALK*-positive NSCLC revealed a response rate of 57% (95% CI: 46% to 68%) and an estimated 6-month PFS probability of 72% (95% CI: 61% to 83%) [10]. A retrospective review of 82 *ALK*-positive patients (including patients who had received multiple lines of therapy) treated with crizotinib revealed an impressive 1-year survival of 74% (95% CI: 63% to 82%) and 2-year survival of 54% (95% CI: 40% to 66%) [11].

Despite the significant improvement in outcomes for these highly selected patients, treatment failures secondary to resistance are now being seen. Combinations of targeted therapies or dual inhibitors may overcome resistance; however, it is more likely that such strategies will simply delay the inevitable. The subsections that follow review the potential targets that hold promise for NSCLC patients and the current or planned trials involving those targets. Table I summarizes what we feel are the relevant trials at the time of writing.

2. EGFR INHIBITION

Dysregulation of EGFR is associated with poorer prognosis in NSCLC [11]. Although many patients with *EGFR* mutation will benefit from EGFR-TKIs, it is now appreciated a portion of the population will not benefit and that some will acquire resistance to those agents. Known mechanisms of resistance include an *EGFR* mutation that reduces the inhibitory ability of gefitinib or erlotinib, and *MET* amplification with subsequent activation of downstream pathways [12,13].

The discovery of resistance to the EGFR-TKIs has led to the development of second-generation EGFR-TKIs. PF 0299804 (dacomitinib) is currently the subject of a phase III trial that will compare it with placebo in patients in whom standard treatment has failed.

PF 0299804 is an oral irreversible inhibitor of the EGFR/HER1, HER2, and HER4 tyrosine kinases. Preclinical data showed activity for PF 0299804 against *EGFR* mutations and T790M [12,14]. Two phase II studies highlighted the agent's clinical antitumour effect, both in first-line therapy and in treatment-refractory patients. In the first of the studies, PF 0299804 was compared with erlotinib. That trial enrolled a range of molecular subgroups, including a group of patients with wild-type *KRAS*. In all subgroups, PF 0299804 showed a PFS advantage (12.4 weeks vs. 8.3 weeks; HR: 0.704; $p = 0.030$) [15]. Another phase II study of PF 0299804 showed preliminary evidence of tumour shrinkage in patients with *EGFR* activating mutations, wild-type *EGFR,* and mutations in exon 20 [16].

Afatinib, an irreversible EGFR-, HER2-, and HER4-TKI demonstrated preclinical activity against the T790M mutation. The phase IIB/III LUX-Lung 1 trial (randomized, double-blind) examined best supportive care plus afatinib or placebo in patients in whom chemotherapy and a reversible EGFR inhibitor had failed. No difference in overall survival was observed; however, PFS was significantly improved with afatinib (3.3 months vs. 1.1 months with placebo; HR: 0.38; 95% CI: 0.306 to 0.475; $p < 0.001$), as were tumour-related symptoms [17] and quality of life.

A number of phase II trials are currently examining afatinib. NCT00525148 has completed enrolment of patients with activating *EGFR* mutations in whom first-line chemotherapy has failed. Similarly, NCT00711594, a Japanese trial, has completed accrual; results are awaited from this group of stage IIIB/IV NSCLC patients who progressed after platinum chemotherapy and either erlotinib or gefitinib. A phase I trial, NCT01169675 will assess afatinib in combination with pemetrexed. The LUX-Lung 3 trial has finished accrual of patients with *EGFR* mutation–positive tumours who are being treated in the first line with afatinib or cisplatin and pemetrexed. Results are to be reported at the 2012 American Society of Clinical Oncology annual meeting.

Targeted therapies in combination may have a synergistic benefit. A number of phase I trials are currently examining newer therapies in combination with erlotinib. Cetuximab, pazopanib, BKM120 (an inhibitor of PI3K), OSI-906 (an inhibitor of insulin-like growth factor 1 receptor), dovitinib (an inhibitor of fibroblast growth factor receptor), and MM121 (anti-ErbB3) are all in trial in combination with erlotinib.

3. Mek INHIBITION

The Raf/Ras/Mek pathway is one of the key signalling pathways involved in the pathogenesis of lung cancer. Activation of Ras and subsequent recruitment of Raf leads to a cascade of events, including phosphorylation of Mek1/Mek2 and the MAPKs [18]. This pathway creates a number of treatment targets, including the Ras family, BRAF and Mek, with novel therapies already in trial.

K-ras is a member of the Ras family, and *KRAS* mutations are present in 15%–25% of NSCLC tumours [19]. The importance of *KRAS* as a prognostic marker arose from patient treatment failures in trials of EGFR inhibitors. A meta-analysis examined the ability of *KRAS* mutations to predict a lack of response to EGFR targeting agents. The sensitivity was low (0.47; 95% CI: 0.43 to 0.52), indicating alternative mechanisms

TABLE I Current or planned trials involving targeted agents relevant to patients with non-small-cell lung cancer

Targeted agent	*ClinicalTrials.gov ID*	*Phase*	*Status*	*Population*	*Estimated enrollment (n)*	*Estimated completion date*	*Trial site*
EGFR inhibition							
Erlotinib vs. docetaxel	NCT00637910	III	Recruiting	Stage IIIB/IV NSCLC, wild-type *EGFR,* prior platinum chemotherapy, no prior taxanes	850	Feb 2013	Italy
Erlotinib + cetuximab	NCT00397384	I/II	Active, not recruiting	Advanced malignancy: head-and-neck, gastrointestinal tract, NSCLC, colorectal	49	Jan 2013	U.S.A.
Erlotinib + pazopanib	NCT01027598	II	Active, not recruiting	Stage IIIB/IV NSCLC, 1 prior chemotherapy	90	Dec 2011	U.S.A.
Erlotinib + BKM120	NCT01487265	I/II	Not yet recruiting	Progressive NSCLC, prior chemotherapy, sensitivity to EGFR-TKIS	73	Oct 2014	U.S.A.
Erlotinib + OSI-906	NCT01221077	II	Recruiting	Stage IIIB/IV NSCLC, *EGFR* mutation, chemotherapy-naïve	86	Jul 2014	Int
Erlotinib + ARQ 197 (tivantinib)	NCT01377376	III	Recruiting	Stage IIIB/IV NSCLC, wild-type *EGFR,* prior platinum chemotherapy	—	Dec 2013	Japan
Erlotinib + ARQ 197 (tivantinib)	NCT01244191	III	Recruiting	Stage IIIB/IV NSCLC, 2 prior lines of chemotherapy	988	Jul 2013	Int
Erlotinib + dovitinib	NCT01515969	I	Not yet recruiting	Stage IIIB/IV NSCLC, prior treatment	30	Dec 2013	U.S.A.
Erlotinib + hydroxychloroquine	NCT00977470	II	Recruiting	Stage IV NSCLC, *EGFR* positive, no prior treatment	76	Aug 2013	U.S.A.

TABLE I Current or planned trials involving targeted agents relevant to patients with non-small-cell lung cancer *(Continued)*

Targeted agent	*ClinicalTrials.gov ID*	*Phase*	*Status*	*Population*	*Estimated enrollment (n)*	*Estimated completion date*	*Trial site*
Carboplatin, paclitaxel, bevacizumab ± erlotinib	NCT00976677	II	Active, not recruiting	Stage III/IV NSCLC, non-squamous, nonsmokers	189	Jul 2012	U.S.A.
Gefitinib	NCT01203917	IV	Recruiting	Stage IIIB/IV NSCLC, Caucasian, *EGFR* mutation	100	Aug 2012	Eur
Gefitinib (maintenance)	NCT01404260	III	Active, not recruiting	Stage IIIB/IV NCSLC, stable disease after chemotherapy, *EGFR* unknown, never or light smokers	218	Apr 2014	China
Gefitinib (rechallenge)	NCT01530334	II	Not yet recruiting	Previous response to gefitinib, subsequent chemotherapy	92	Sep 2013	Italy
Gefitinib vs. pemetrexed	NCT00891579	II	Recruiting	Stage IIIB/IV NSCLC, wild-type *EGFR,* prior platinum chemotherapy	150	May 2012	China
AP26113	NCT01449461	I/II	Recruiting	Advanced/metastatic malignancy	130	Sep 2015	U.S.A.
CO-1686	NCT01526928	I/II	Not yet recruiting	Stage IIIB/IV NSCLC, *EGFR* mutation	70	May 2014	U.S.A.
Afatinib	NCT00525148	II	Active, not recruiting	Stage IIIB/IV NSCLC, *EGFR* mutation	120	Jul 2012	Int
Afatinib	NCT00711594	II	Active, not recruiting	Stage IIIB/IV NSCLC, prior platinum chemotherapy, progressed after gefitinib or erlotinib	72	Dec 2012	Japan

TABLE I Current or planned trials involving targeted agents relevant to patients with non-small-cell lung cancer *(Continued)*

Targeted agent	*ClinicalTrials.gov ID*	*Phase*	*Status*	*Population*	*Estimated enrollment (n)*	*Estimated completion date*	*Trial site*
Afatinib	NCT01542437	II	Recruiting	Stage IIIB/IV NSCLC, at least 1 prior chemotherapy	150	Dec 2012	Mexico
Afatinib + pemetrexed	NCT01169675	I	Recruiting	Advanced solid tumours	90	May 2012	Canada
PF-00299804	NCT01000025	III	Recruiting	Stage IIIB/IV NSCLC	720	Nov 2012	Int
MM121 + erlotinib	NCT00994123	I/II	Recruiting	Stage IIIB/IV NSCLC	260	Feb 2013	U.S.A.
MM121 + irinotecan + cetuximab	NCT01451632	I	Recruiting	Advanced malignancy, no standard options remaining	45	Oct 2013	U.S.A.
Afatinib + sirolimus	NCT00993499	I	Recruiting	Stage IIIB/IV NSCLC, *EGFR* mutation or progression after erlotinib in *EGFR* wild-type	42	Aug 2014	Spain
BRAF inhibition							
AZD6244	NCT01306045	II	Recruiting	Stage IV NSCLC, SCLC, and thymic cancer	600	Jan 2017	U.S.A.
AZD6244	NCT00888134	I	Active, not recruiting	*BRAF*-mutated malignancy	66	Apr 2012	U.S.A.
Dasatinib	NCT01514864	II	Not yet recruiting	Stage IV NSCLC or melanoma, with *DDR2* mutation or *BRAF* mutation	95	Mar 2017	Int
Akt inhibition							
MK-2206 + erlotinib	NCT01294306	II	Recruiting	Advanced NSCLC, progressed after initial response to erlotinib	90	Nov 2011	U.S.A.

TABLE I Current or planned trials involving targeted agents relevant to patients with non-small-cell lung cancer *(Continued)*

Targeted agent	*ClinicalTrials.gov ID*	*Phase*	*Status*	*Population*	*Estimated enrollment (n)*	*Estimated completion date*	*Trial site*
MK-2206 + gefitinib	NCT01147211	I	Recruiting	Advanced NSCLC, prior EGFR inhibitor, prior platinum chemotherapy	21	Dec 2012	Taiwan
PI3K inhibition							
BYL719 + MEK162	NCT01449058	I/II	Not yet recruiting	Advanced incurable tumour	58	Mar 2014	Int
XL147 + erlotinib	NCT00692640	I	Active, not recruiting	Advanced incurable tumour	65	Nov 2011	U.S.A.
Mek1 inhibition							
GSK2118436	NCT01362296	II	Recruiting	Stage IV NSCLC, positive mutational status for *KRAS, BRAF, NRAS, MEK1,* 1 prior platinum treatment	141	Aug 2012	Int
BKM120 + MEK162	NCT01363232	I/II	Recruiting	Advanced incurable tumour	58	Jul 2012	Int
BEZ235 +MEK162	NCT01337765	I/II	Recruiting	Advanced incurable tumour	55	Jun 2013	Int
GSK1120212 + gemcitabine	NCT01324258	I	Recruiting	Advanced incurable tumour, Japanese patients	19	Jun 2012	Japan
GSK1120212 + BKM120	NCT01155453	I	Recruiting	Advanced incurable tumour	60	Jul 2013	Int
HDAC inhibition							
LBH589 + erlotinib	NCT00738751	I	Not yet recruiting	Stage IV NSCLC or stage IV head-and-neck cancer	44	Dec 2012	U.S.A.
Vorinostat + hydroxychloroquine	NCT01023737	I	Recruiting	Advanced incurable tumour	30	Nov 2012	U.S.A.

TABLE I Current or planned trials involving targeted agents relevant to patients with non-small-cell lung cancer *(Continued)*

Targeted agent	*ClinicalTrials.gov ID*	*Phase*	*Status*	*Population*	*Estimated enrollment (n)*	*Estimated completion date*	*Trial site*
Vorinostat + NPI-0052	NCT00667082	I	Recruiting	Advanced/metastatic NSCLC, pancreatic cancer, melanoma, lymphoma	40	Sep 2009	Aus
CUDC-101	NCT01171924	IB	Active, not recruiting	Stage IV NSCLC with *EGFR* mutation (exons 19/21), progressed with erlotinib, breast, gastric, head-and-neck	40	Oct 2011	U.S.A.
Vorinostat + sirolimus	NCT01087554	I	Recruiting	Advanced incurable biopsy-proven solid tumour	65	Mar 2014	U.S.A.
Vorinostat or sirolimus + hydroxychloroquine	NCT01266057	I	Recruiting	Advanced or metastatic malignancy, failed treatment	160	Apr 2015	U.S.A.
Vorinostat + gefitinib	NCT01027676	II/III	Recruiting	Stage IIIB/IV NSCLC, 1 platinum-based chemotherapy	50	Dec 2012	Korea
Vorinostat + bortezomib	NCT00798720	II	Completed recruiting	Advanced NSCLC, 2 prior chemotherapy regimens	33	Dec 2011	U.S.A.
Entinostat + sorafenib	NCT01159301	I	Recruiting	Advanced incurable tumour	44	May 2011	U.S.A.
PXD101 (belinostat) + bevacizumab + carboplatin + paclitaxel	NCT01090830	I/II	Recruiting	Stage IV NSCLC, no previous treatment	28	Sep 2012	U.S.A.
LBH589 + pemetrexed	NCT00907179	I/II	Recruiting	Stage IV NSCLC, 1 prior chemotherapy regime	50	Jun 2013	U.S.A.
Vorinostat + pazopanib	NCT01339871	I	Recruiting	Advanced incurable tumour	134	Apr 2016	U.S.A.

TABLE I Current or planned trials involving targeted agents relevant to patients with non-small-cell lung cancer *(Continued)*

Targeted agent	*ClinicalTrials.gov ID*	*Phase*	*Status*	*Population*	*Estimated enrollment (n)*	*Estimated completion date*	*Trial site*
Alk inhibition							
Crizotinib (PF-02341066)	NCT01500824	II	Not yet recruiting	Stage IV NSCLC, non-squamous, East Asian, with *ALK* fusion gene, 1 prior chemotherapy	50	Feb 2014	China
Crizotinib (PF-02341066)	NCT00932451	II	Recruiting	Stage IV NSCLC, with *ALK* fusion gene	100	Mar 2013	Int
Crizotinib (PF-02341066) + PF-00299804 (pan-HER inhibitor)	NCT01121575	I	Recruiting	Stage IV NSCLC, acquired resistance to gefitinib and erlotinib	70	Nov 2012	Int
Crizotinib (PF-02341066) + PF-00299804 (pan-HER inhibitor)	NCT01441128	I	Recruiting	Stage IV NSCLC	22	Nov 2016	U.S.A.
Crizotinib (PF-02341066) vs. pemetrexed or docetaxel	NCT00932893	III	Recruiting	Stage IV NSCLC, with *ALK* fusion gene	318	Feb 2013	Int
LDK378	NCT01283516	I	Recruiting	Stage IV NSCLC, with *ALK* fusion gene	70	Jul 2013	Int
AP26113	NCT01449461	I/II	Recruiting	Stage IV NSCLC	130	Sep 2015	U.S.A.
CMET inhibition							
PF-02341066 (CMET inhibitor) + PF-00299804 (pan-HER inhibitor)	NCT01121575	I	Recruiting	Advanced NSCLC, resistance to erlotinib and gefitinib	70	Nov 2012	Int
KRAS *mutations*							
AZD6244 + erlotinib	NCT01229150	II	Recruiting	Advanced NSCLC, 1 prior platinum chemotherapy	100	Sep 2014	U.S.A.

TABLE I Current or planned trials involving targeted agents relevant to patients with non-small-cell lung cancer *(Continued)*

Targeted agent	*ClinicalTrials.gov ID*	*Phase*	*Status*	*Population*	*Estimated enrollment (n)*	*Estimated completion date*	*Trial site*
Erlotinib + ARQ 197 vs. single-agent chemotherapy	NCT01395758	II	Recruiting	Stage IV NSCLC, with *KRAS* mutation	98	Jun 2012	U.S.A.
GSK1120212 vs. docetaxel	NCT01362296	II	Recruiting	Stage IV NSCLC, positive mutational status for *KRAS, NRAS, BRAF, MEK1*	141	Aug 2012	Int

NSCLC = non-small-cell lung cancer; EGFR-TKI = epidermal growth factor receptor tyrosine kinase inhibitor; Int = international (more than one continent); Eur = Europe; SCLC = small-cell lung cancer; Aus = Australia; HER = human epidermal growth factor receptor.

for resistance to EGFR inhibitors. However, *KRAS* was found to have a high specificity, suggesting that patients with a *KRAS* mutation were unlikely to respond to treatment based on anti-EGFR agents [20]. That finding has led to stratification and prognostication based on the *KRAS* mutation status of patients.

Although K-ras is difficult to target, interest in Mek inhibition is increasing. As a downstream effector of the EGFR pathway that signals through K-ras, Mek inhibition may play a role in patients who become resistant to EGFR inhibitors. A number of trials to examine Mek inhibitors alone or in combination with other targeted treatments are currently recruiting. GSK2118436 is a potent Mek inhibitor that has been shown to have preclinical activity in NSCLC and melanoma. A phase II trial is currently recruiting and will examine GSK2118436 in comparison with docetaxel in advanced NSCLC patients with a *KRAS* mutation. For exploratory purposes, a small population of patients with *BRAF, NRAS,* and *MEK1* mutations will also be included. Patients must have had previous exposure to platinum chemotherapy. The primary outcome will be PFS, and the trial is expected to be completed in late 2012.

A phase I trial of GSK1120212, another potent Mek inhibitor, in combination with gemcitabine is currently recruiting in Japan. MEK162 is a Mek1/2 inhibitor, and a number of phase I trials are currently examining the combination of this Mek inhibitor with PI3K inhibitors.

4. BRAF INHIBITION

The protein kinase BRAF is a member of the Raf family, kinases that act downstream of Ras and are responsible for further activation of the MAPK pathway, controlling cellular proliferation [21]. *BRAF* mutations were first identified in melanoma cells, with 80% of mutations involving the Val600 residue in the kinase domain. By contrast, *BRAF* mutations account for only 1%–3% of NSCLC. These non-Val600Glu mutations include Gly468Ala and Leu596Val [22–24]. *BRAF* mutations result in increased kinase activity and activation of MAPK2 and MAPK3 [24]. A number of studies are currently examining the effect of therapeutic agents on *BRAF*-mutated patients. AZD6244 is a Mek inhibitor that is the focus of two trials specifically examining its effect on *BRAF*-mutated patients. The NCT00888134 phase II trial of AZD6244 is enrolling patients with metastatic malignancy and a *BRAF* mutation. The primary outcome is response rate, with secondary outcomes of PFS and response rate specifically in the NSCLC and colon cancer populations.

NCT01514864, although not yet recruiting, is planning to examine the effect of dasatinib in patients with a malignancy harbouring a *BRAF* mutation. Dasatinib is a Src-TKI, and abnormal expression of activated Src has been found in many tumour types, including NSCLC [25]. High levels of Src expression and

activation have been correlated with poor prognosis in human malignancies [26]. Src has been shown to interact with many signalling pathways, including EGFR, MAPK, PI3K/Akt, and vascular endothelial growth factor [27]. The role of Src as a therapeutic target in NSCLC is supported by preclinical studies [28]. NCT01514864 will include patients with NSCLC and melanoma with *BRAF* mutations, squamous cell NSCLC with a *DDR2* mutation, and any other malignancies with one of those two mutations.

5. Alk INHIBITION

Knowledge about *ALK* gene rearrangements, including the *EML4–ALK* fusion, is rapidly growing. That fusion arises from an inversion on the short arm of chromosome 2 that joins exons 1–14 of *EML4* with exons 20–29 of *ALK* [29]. Despite having a phenotype similar to that seen in *EGFR* mutations, *ALK* mutations occur almost exclusively in the absence of *EGFR* and *KRAS* mutations. Preclinical trials have shown that cells habouring *ALK* mutations are exquisitely sensitive to Alk inhibition [30]. Crizotinib was the first of the Alk inhibitors to gain profile. The impressive results of a phase I trial, with a response rate of 57% and an estimated 6-month PFS of 72%, led directly to a phase III trial and a host of other trials aiming to clarify the role of crizotinib in treatment [31].

The phase III trial is comparing crizotinib with single-agent chemotherapy after a platinum-based regimen in 318 patients with a mutated *ALK* gene. The primary outcome is PFS, with secondary outcomes including response rate, overall survival, patient-reported symptoms, and *EML4–ALK* variants.

The exact position of crizotinib in the treatment paradigm continues to be explored in trials in the first line of treatment that are currently recruiting. Crizotinib pharmacokinetics have already been identified to possibly be different in Asian populations [10], but to date, that hypothesis has not been clarified. The phase II NCT01500824 trial, which will focus on previously-treated *ALK*-mutated East Asian patients is not yet open to recruitment. The primary outcome will be response rate.

Investigation of resistance to current EGFR inhibitors has highlighted the role of the c-Met/Alk pathway. Resistance can occur through not only the EGFR pathway, but also c-Met/Alk. Combinations of EGFR and c-Met/Alk inhibitors therefore hold potential for overcoming resistance [32]. PF 0299804 is an irreversible pan-HER inhibitor. Preclinical data have shown that this agent has activity against *EGFR* activating mutations and T790M [13,33]. Two trials, currently recruiting, will examine the combination of crizotinib and PF 0299804, studying the combination in patients who show resistance to the EGFR inhibitor (gefitinib or erlotinib).

New Alk inhibitors are under investigation, with phase I trials of LDK378 and AP26113 currently recruiting. NCT01449461, a phase I trial of AP26113, will be conducted in two parts, with the second part including expansion cohorts. The cohorts include *ALK* mutations with no previous exposure to Alk inhibitors, *ALK* mutation with resistance to an Alk inhibitor, *EGFR* mutation with resistance to EGFR inhibitors, and non-lung malignancies with *ALK* mutations.

6. HDAC INHIBITION

Modification of DNA and histones is the most common method of epigenetic gene control. The family of histone deacetylases (HDACS) plays a central role in that process. The HDACS act to tighten the bond between histones and DNA, thus inhibiting gene transcription by blocking binding sites on promoters [15]. Inhibition of HDACS leads to induction of apoptosis in malignant cells [34]. The HDAC inhibitors continue to be investigated in NSCLC, and preclinical data suggest that they may increase E-cadherin and sensitize malignant cells to EGFR inhibition [35]. Consequently, in addition to monotherapy trials of new HDAC inhibitors, combinations of EGFR-TKIS with HDAC inhibitors are receiving focus.

In this class of therapeutics, vorinostat is currently the furthest along in development. Vorinostat is already known to act synergistically with a variety of chemotherapy agents, suggesting that it works through number of pathways, shifting the balance of apoptotic genes, inducing reactive oxygen species, and inhibiting angiogenesis [36]. In preclinical studies, vorinostat combined with platinum or taxanes exhibits synergistic antitumour effects [37,38]. A phase II/III trial in combination with paclitaxel and carboplatin was unfortunately stopped early because the trial did not meet a pre-specified proof-of-concept criterion.

A number of phase I trials to examine the effect of vorinostat with other targeted treatments are currently recruiting. Those targeted treatments include inhibitors of EGFR, vascular endothelial growth factor, the mammalian target of rapamycin (MTOR), and a proteasome inhibitor, NP10052. Vorinostat is well tolerated, a clinical advantage when looking for combination therapies.

New HDAC inhibitors continue to be investigated. The effects of LBH589 (panobinostat) are currently under examination in both hematologic and solid tumours. Preclinical studies show synergy not only with chemotherapy, but also with proteasome inhibitors and demethylators [39,40]. Two phase I trials of LDH589 are combining it with pemetrexed and erlotinib chemotherapy respectively. Belinostat (PXD101), another HDAC inhibitor, has shown efficacy for induction of apoptosis in a number of solid malignancies [41]. A phase I/II study, NCT01090830, currently recruiting, will examine belinostat in combination with bevacizumab and platinum chemotherapy in treatment-naïve patients.

7. PI3K/Akt INHIBITION

The PI3K/Akt/mTOR pathway is essential for cell proliferation, protein synthesis, and angiogenesis. The PI3K/Akt pathway upregulates mTOR in response to stimulation by growth factors [42]. The tumour suppressor gene *PTEN* antagonizes the PI3K/Akt pathway. Loss of inactivating mutations of *PTEN* results in a gain in function of the *PI3KCA* gene. Loss of *PTEN,* resulting in overexpression of phosphorylated Akt, is associated with poorer prognosis in lung cancer [43].

Several small molecules in early-phase clinical trials are currently known to target the mTOR pathway. Everolimus, an oral mTOR inhibitor, has shown activity in metastatic NSCLC. A phase II study of everolimus examined its use in NSCLC patients who had received prior chemotherapy or erlotinib. The median PFS was 2.6 months, and the relative risk, 4.6% [44]. Even in the absence of the *PIK3CA* mutation, the mTOR inhibitors may be active, because dysregulation of mTOR occurs at several levels. Preclinical trials of PI3K inhibitors have shown efficacy, and research is ongoing [45,46]. BYL719 is a selective inhibitor of PI3Kα. A phase I/II trial will see it combined with the Mek inhibitor MEK162. This international multicentre trial is not yet recruiting, but is expected to be completed by 2014.

8. NOVEL THERAPIES

In the current environment of targeted treatments, oncolytic viruses are also attracting increasing attention. Reolysin (Oncolytics Biotech, Calgary, AB) is a clinical GMP strain of reovirus serotype 3–Dearing. It is a double-stranded RNA virus that specifically replicates in cells possessing an activated Ras signalling pathway (or up- and downstream elements of that pathway) [47,48]. Preclinical data highlight the ability of reovirus to repetitively replicate within cells and demonstrated a synergistic effect with chemotherapy (especially with microtubule-inhibiting agents) and radiation [49,50]. There are data to suggest that the anticancer effects of Reolysin may be augmented by combination with chemotherapy directly and by the potential of chemotherapy to reduce immune clearance of the reovirus [51,52].

Reolysin is the focus of a number of clinical trials involving solid tumours. A phase II study, NCT00861627, is currently recruiting patients. It will examine Reolysin in combination with carboplatin and paclitaxel. The trial aims to enroll 36 patients with stage IIIB/IV NSCLC having a *KRAS* or *EGFR* activating mutation. The patients will be chemonaïve, but may have received treatment with an EGFR-TKI.

9. SUMMARY

The discovery of new biomarkers for targeted therapies has greatly changed the management and prognosis of many patients with NSCLC. Further, knowledge of the molecular pathways and mutational drivers of lung cancer will expand the use of targeted treatments. Hopefully, the identification of new therapeutic targets such as Alk, c-Met, mTOR, and PI3K, and investigations into simultaneously inhibiting multiple pathways and overcoming resistance, will provide personalized and precise treatments for lung cancer patients in the near future.

10. CONFLICT OF INTEREST DISCLOSURES

The authors have no financial conflicts of interest to declare.

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